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				1	/54	 -			
Activity	U/g IB	0	0	0	40.00	0	105.26	15.65	48.70
550 mM	L- Arg	0	+	0	+	0	+	0	+
440 mM	Sucrose	0	+	+	0	+	0	0	+
2.2 mM	CaCl ₂	+	0	+	0	0	+	0	+
2.2 mM	MgCl ₂	+	0	+	0	0	+	0	4
1.1 mM	EDTA	0	+	- 0	+	+	0	+	0
550 mM	GndHCl	+	+	0	0	0	0	+	+
0.055%	PEG 3350	0	+	0	+	0	+	0	+
Mm	KCI	0.44	0.44	11	11	0.44	0.44	11	11
Mm	NaCl	10.56	10.56	264	264	10.56	10.56	264	264
0.3 mM	LM	+	0	0	+	+	, 0	0	+
0.1 mM	GSSG	+	+	+	+	+	+	+	+
1 mM	HSS	+	+	+	+	+	+	+	+
	#	2 (55 mM MES pH 6.5)	3 (55 mM MES pH 6.5)	#5 (55 mM MES pH 6.5)	#8 (55 mM MES pH 6.5)	#10 (55 mM MES pH 8.2)	#11 (55 mM Tris pH 8.2)	#13 (55 mM Tris pH 8.2)	#16 (55 mM Tris pH 8.2)

ELution of MBP-ST3Gal III from Amylose column

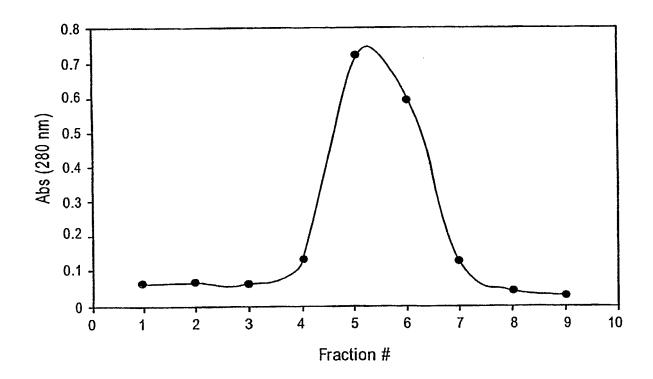


FIG. 2

ST3 Gal III activities of the Amylose purified refolded MBP-ST3Gal III fractions

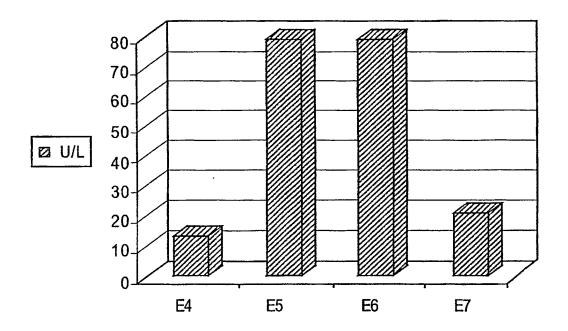


FIG. 3

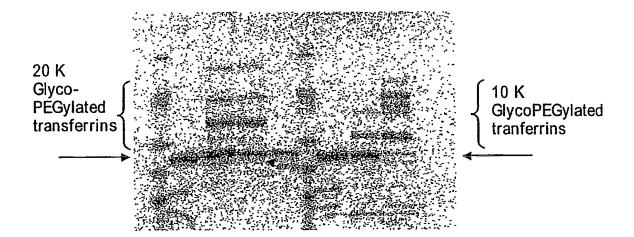
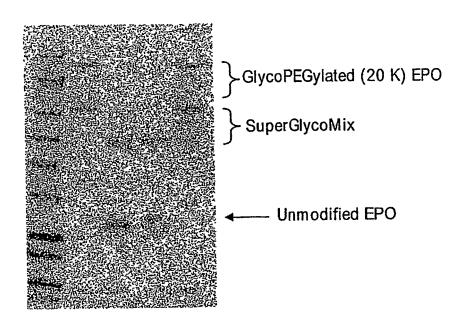


FIG. 4



GlycoPEGylation (20 K) of EPO

FIG. 5

/ /	Net retroop	10	20	30		50 GDPASLTREV	00 COA.TST
/usr/t	MILKKOSA	AGLVLWGALL	T VAWWATITI	FEWIRPARG	KEESVSALLX	TOPASHINEV.	·····
D27115	MI KKUS	ACT VI WCATT	I.T.TANWAYA.	FFWTRPVPS		DDPASLTREV	
FZ /113	IATTA (NOV	10	20	30	40	50	60
		10	20	30			
		70		90	100	110	
/usr/t	AEVELE	RORGLIQOIO	DALSSQRO	RVPTAAPPA	QPRVPVTPA	PAVIPILVIA	CDRSTV
	::::::	::::::::	. :: :::	.::::::::	::.::::	::::::::	::::::
P27115	AEVELE	RQRGLLQQIF	REHHALWSQRU	VKVPTAAPPA	QPHVPVTPP	PAVIPILVIA	CDRSTV
		70	80	90	100	110	120
				450	1.00	170	
	120		140	150	160	170	אבוו זכונו
/usr/t	RRCLDK	LLHYRPSAEI				RQPDLSSIAV	
	::::::	***********				CONTRACTOR	
P27115	RRCLDK	LLHYRPSAEI	TELTIASÓINO	3HEETAQVIA 150	160	RQPDLSNIAV 170	180
		130	140	130	100	170	100
	180	190	200	210	220	230	
/ner/t	EUGAAK TOO					RATYPLLKAD	PSLWCV
			::.:: .:	:::::::::::	:::::::::::::::::::::::::::::::::::::::	.::::::::	::::::
P27115	FOGYYK	(TARHYRWAL	GOIFHNFNYP	AAVVVEDDLI	EVAPDFFEYF	QATYPLLKAL	PSLWCV
12/11	- 20	190	200	210	220	230	240
	240	250	260			290	
/usr/t	240 SAWNDIN	250 IGKEQMVDAS				WPKAFWDDWM	
	SAWNDN	IGKEQMVDAS	RPELLYRTDF	FPGLGWLLL	AELWAELEPK	WPKAFWDDW	::::::
	SAWNDN	IGKEQMVDAS IGKEQMVDSS	RPELLYRTDF .:::::: KPELLYRTDF	FPGLGWLLLA FPGLGWLLLA	AELWAELEPK :::::::: AELWAELEPK	WPKAFWDDW WPKAFWDDW	IRRPEQR
	SAWNDN	IGKEQMVDAS	RPELLYRTDF	FPGLGWLLL	AELWAELEPK	WPKAFWDDW	::::::
	SAWNDN	IGKEQMVDAS :::::::::: IGKEQMVDSS 250	RPELLYRTDF .::::::: KPELLYRTDF 260	FPGLGWLLLZ ::::::: FPGLGWLLLZ 270	AELWAELEPK :::::::: AELWAELEPK 280	WPKAFWDDWN WPKAFWDDWN 290	IRRPEQR
P27115	SAWNDN SAWNDN	IGKEQMVDAS ::::::::: IGKEQMVDSS 250 310	RPELLYRTDF .:::::: KPELLYRTDF 260 320	FPGLGWLLLZ :::::::: FPGLGWLLLZ 270 330	AELWAELEPK :::::::: AELWAELEPK 280 340	WPKAFWDDWN WPKAFWDDWN 290 350	IRRPEQR 300
P27115	SAWNDN SAWNDN	IGKEQMVDAS ::::::::: IGKEQMVDSS 250 310 IRPEISRTMT	RPELLYRTDF .:::::: KPELLYRTDF 260 320 FGRKGVSHGQ	FPGLGWLLLA FPGLGWLLLA 270 330 FFDQHLKFT	AELWAELEPK :::::::: AELWAELEPK 280 340 KLNQQFVHFT	WPKAFWDDWN WPKAFWDDWN 290	IRRPEQR 300
P27115	SAWNDN SAWNDN 300 QGRACI	IGKEQMVDAS IGKEQMVDSS 250 310 IRPEISRTMT	RPELLYRTDF .::::::: KPELLYRTDF 260 320 FGRKGVSHGQ	FPGLGWLLLZ FPGLGWLLLZ 270 330 FFDQHLKFT	AELWAELEPK AELWAELEPK 280 340 KLNQQFVHFT	WPKAFWDDWN WPKAFWDDWN 290 350 PQLDLSYLQRE	IRRPEQR 300 EAYDRDF
P27115	SAWNDN SAWNDN 300 QGRACI	IGKEQMVDAS IGKEQMVDSS 250 310 IRPEISRTMT	RPELLYRTDF .::::::::::::::::::::::::::::::::::::	FPGLGWLLLZ FPGLGWLLLZ 270 330 FFDQHLKFT	AELWAELEPK AELWAELEPK 280 340 KLNQQFVHFT	WPKAFWDDWN WPKAFWDDWN 290 350	IRRPEQR 300 EAYDRDF
P27115	SAWNDN SAWNDN 300 QGRACI	IGKEQMVDAS IGKEQMVDSS 250 310 IRPEISRTMT	RPELLYRTDF .::::::: KPELLYRTDF 260 320 FGRKGVSHGQ	FPGLGWLLLA FPGLGWLLLA 270 330 FFDQHLKFT FFDQHLKFT	AELWAELEPK ELWAELEPK 280 340 KINQQFVHFT	WPKAFWDDWN WPKAFWDDWN 290 350 CQLDLSYLQRE	IRRPEQR 300 EAYDRDF EAYDRDF
P27115 /usr/t	SAWNDN SAWNDN SAWNDN SOR	IGKEQMVDAS IGKEQMVDSS 250 310 IRPEISRIMI VRPEISRIMI 310 370	RPELLYRTDF .::::::: KPELLYRTDF 260 320 FGRKGVSHCQ :::::::: FGRKGVSHGQ 320 380	FPGLGWLLLZ FPGLGWLLLZ 270 330 FFDQHLKFT 330 390	AELWAELEPK :::::::: AELWAELEPK 280 340 KLNQQFVHFT :::::::::: KLNQQFVPFT 340 400	WPKAFWDDWN 290 350 QLDLSYLQRE QLDLSYLQRE 350 410	ARRPEQR 300 CAYDRDF CAYDRDF 360
P27115 /usr/t	SAWNDN SAWNDN SAWNDN SOR	IGKEQMVDAS IGKEQMVDSS 250 310 IRPEISRIMI VRPEISRIMI 310 370	RPELLYRTDF .::::::: KPELLYRTDF 260 320 FGRKGVSHCQ :::::::: FGRKGVSHGQ 320 380	FPGLGWLLLZ FPGLGWLLLZ 270 330 FFDQHLKFT 330 390	AELWAELEPK :::::::: AELWAELEPK 280 340 KLNQQFVHFT :::::::::: KLNQQFVPFT 340 400	WPKAFWDDWN 290 350 PQLDLSYLQRE PQLDLSYLQRE 350	ARRPEQR 300 CAYDRDF CAYDRDF 360
/usr/t P27115	SAWNDN SAWNDN SOO GGRACI KGRACV SOO LARVYO	IGKEQMVDAS 250 310 IRPEISRIMI VRPEISRIMI 310 370 GAPQLQVEKV	RPELLYRTDF .::::::::::::::::::::::::::::::::::::	FPGLGWLLLZ 270 330 FFDQHLKFT 330 390 CVRVQYTGRD	AELWAELEPK 280 340 KLNQQFVHFT 340 400 SFKAFAKALO	WPKAFWDDWN 290 350 PQLDLSYLQRE SOUDLSYLQQE 350 410 SVMDDLKSGVI	IRRPEQR 300 EAYDRDF EAYDRDF 360 PRAGYRG
/usr/t P27115	SAWNDN SAWNDN SOO GGRACI KGRACV SOO LARVYO	IGKEQMVDAS 250 310 IRPEISRIMI VRPEISRIMI 310 370 GAPQLQVEKV	RPELLYRTDF .::::::::::::::::::::::::::::::::::::	FPGLGWLLLZ 270 330 FFDQHLKFT 330 390 CVRVQYTGRD	AELWAELEPK 280 340 KLNQQFVHFT 340 400 SFKAFAKALA	WPKAFWDDWN 290 350 QLDLSYLQRE 350 410 EVMDDLKSGVI	EAYDRDF 360 PRAGYRG PRAGYRG
/usr/t P27115	SAWNDN SAWNDN SOO GGRACI KGRACV SOO LARVYO	IGKEQMVDAS 250 310 IRPEISRIMI VRPEISRIMI 310 370 GAPQLQVEKV	RPELLYRTDF .::::::::::::::::::::::::::::::::::::	FPGLGWLLLZ 270 330 FFDQHLKFT 330 390 CVRVQYTGRD	AELWAELEPK 280 340 KLNQQFVHFT 340 400 SFKAFAKALO	WPKAFWDDWN 290 350 PQLDLSYLQRE SOUDLSYLQQE 350 410 SVMDDLKSGVI	IRRPEQR 300 EAYDRDF EAYDRDF 360 PRAGYRG
/usr/t P27115	SAWNDN SAWND SAWNDN SAWNDN SAWND SAWNDN SAWND	IGKEQMVDAS IGKEQMVDSS 250 310 IRPEISRIMI 310 370 GAPQLQVEKV 370	RPELLYRTDF .::::::: KPELLYRTDF 260 320 FGRKGVSHGQ 320 380 RINDRKELGE 2RINDRKELGE 380	FPGLGWLLLZ 270 330 FFDQHLKFT 330 390 CVRVQYTGRD	AELWAELEPK 280 340 KLNQQFVHFT 340 400 SFKAFAKALA	WPKAFWDDWN 290 350 QLDLSYLQRE 350 410 EVMDDLKSGVI	EAYDRDF 360 PRAGYRG PRAGYRG
/usr/t P27115 /usr/t P27115	300 COGRACI CO	IGKEQMVDAS IGKEQMVDSS 250 310 IRPEISRIMI 310 370 GAPQLQVEKV 370 430	RPELLYRTDF .::::::: KPELLYRTDF 260 320 FGRKGVSHGQ 320 380 RTNDRKELGE 380 ATNDRKELGE 380 440	FPGLGWLLLZ FPGLGWLLLZ 270 330 FFDQHLKFT 330 390 CVRVQYTGRD 390 CVRVQYTGRD 390	AELWAELEPK 280 340 KLNQQFVHFT 340 400 SFKAFAKALA	WPKAFWDDWN 290 350 QLDLSYLQRE 350 410 EVMDDLKSGVI	EAYDRDF 360 PRAGYRG PRAGYRG
/usr/t P27115 /usr/t P27115	300 COGRACI CO	IGKEQMVDAS IGKEQMVDSS 250 310 IRPEISRIMI 310 370 GAPQLQVEKV 370 430	RPELLYRTDF .::::::: KPELLYRTDF 260 320 FGRKGVSHGQ 320 380 RINDRKELGE 2RINDRKELGE 380	FPGLGWLLLZ FPGLGWLLLZ 270 330 FFDQHLKFT 330 390 CVRVQYTGRD 390 CVRVQYTGRD 390	AELWAELEPK 280 340 KLNQQFVHFT 340 400 SFKAFAKALA	WPKAFWDDWN 290 350 QLDLSYLQRE 350 410 EVMDDLKSGVI	EAYDRDF 360 PRAGYRG PRAGYRG
/usr/t P27115 /usr/t P27115 /usr/t	300 QGRACI SAWNDN 300 QGRACI LARVYO 420 LIVIFQ	IGKEQMVDAS 250 310 IRPEISRIMI 310 370 370 GAPQLQVEKV 370 430 FRGRRVHLAE	RPELLYRTDF .::::::::::::::::::::::::::::::::::::	FPGLGWLLLZ FPGLGWLLLZ 270 330 FFDQHLKFTI 330 390 CVRVQYTGRD EVRVQYTGRD 390 SWN SWN	AELWAELEPK 280 340 KLNQQFVHFT 340 400 SFKAFAKALO 400	WPKAFWDDWN 290 350 QLDLSYLQRE 350 410 GVMDDLKSGVI 410	EAYDRDF 360 PRAGYRG PRAGYRG
/usr/t P27115 /usr/t P27115 /usr/t	300 QGRACI SAWNDN 300 QGRACI LARVYO 420 LIVIFQ	IGKEQMVDAS 250 310 IRPEISRIMI 310 370 370 GAPQLQVEKV 370 430 FRGRRVHLAE	RPELLYRTDF .::::::: KPELLYRTDF 260 320 FGRKGVSHGQ 320 380 RTNDRKELGE 380 ATNDRKELGE 380 440	FPGLGWLLLZ FPGLGWLLLZ 270 330 FFDQHLKFTI 330 390 CVRVQYTGRD EVRVQYTGRD 390 SWN SWN	AELWAELEPK 280 340 KLNQQFVHFT 340 400 SFKAFAKALO 400	WPKAFWDDWN 290 350 QLDLSYLQRE 350 410 EVMDDLKSGVI	EAYDRDF 360 PRAGYRG PRAGYRG

gqvfrqfrfpaavvveddlevapdffeyfratypllkadpslwcvsawndngkeqmvdasrpellyrtdffpglgwlllaelwae lepkwpkafwddwmrpegragracirpeisrtmtfgrkgvshgqffdqhlkfiklngqfvhffqldlsylqreaydrdflarvyg avipilviacdrstvrrsldkllhyrpsaelfpiivsqdcgheetaqaiasygsavthirqpdlssiavppdhrkfqgyykiarhyrwa $apqlqvekvrtndrkelgevrvqytgrdsfkafakalgvmddlksgvpragyrgivtfqfpgrrvhlappptwegydpswn^{\ast}$

GnT1 Cys121Ser mutant

cagottcaaggotttcgccaaggctctgggtgtcatggatgaccttaagtcggggggttccgagagctggctaccgggggtattgtcacctt Geggtgatteceatectggteategeetgtgaeegeageaetgtteggege**tet**etagaeaagetgetgeattateggeeeteggetga ccctgagatctcaagaacgatgacctttggccgcaagggtgtgagccacgggcagttctttgaccagcacctcaagtttatcaagctga teeggeageeegaeetgageagtageggtgeegeeggaceacegeaagtteeagggetaetaeaagategegegeeactaeeg ctgggcgctgggccaggtcttccggcagtttcgcttcccgcgggcgtggtggtggtggaggatgacctggaggtggccccggacttctt accagcagtttgtgcacttcacccagctggacctgtcttacctgcagcgggaggcctatgaccgagatttcctcgcccgcgtctacggt ccagttcccgggccgccgtgtccacctggcgcccccaccgacgtgggagggctatgatcctagctggaattag

FIG. .

GnT1 Cys121Asp

avipilviacdrstvrrdldkllhyrpsaelfpiivsqdcgheetaqaiasygsavthirqpdlssiavppdhrkfqgyykiarhyrwa lgqvfrqfrfpaavvveddlevapdffeyfratypllkadpslwcvsawndngkeqmvdasrpellyrtdffpglgwlllaelwae lepkwpk afwddwn nr peqrqgrac ir peisrtmt fgrkgv shgqffdqhlk fikln qqfvhffqldl sylqreaydrd flarvyg ${
m apqlq}$ vekvr ${
m tndr}$ kelgev ${
m tvq}$ yt ${
m grdsf}$ kafakal ${
m gvmddlk}$ s ${
m gvpragyrgivtfqfpgrrvhlappptwegydpswn}^*$

agetetteeceateategttagecaggactgegggeacgaggagagaeggeceaggecategeetectaeggeageggeggteaegeae gccctgagatctcaagaacgatgacctttggccgcaagggtgtgagccacgggcagttctttgaccagcacctcaagtttatcaagctg cagettcaaggetttcgccaaggetctggggtgtcatggatgacettaagtcggggggttccgagagctggctaccgggggtattgtcacett aaccagcagtttgtgcacttcacccagctggacctgtcttacctgcagcgggaggcctatgaccgagatttcctcgcccgcgtctacgg gctgggcgctgggccaggtcttccggcagtttcgcttcccggggccgtggtggtggtggaggatgacctggaggtggccccggacttct **Geggtgatteceatectggteategeetgtgacegeageactgtteggegegatetagaeaagetgetgeattateggeeeteggetg** ccagttcccgggccgccgtgtccacctggcgccccaccgacgtgggaggggtatgatcctagctggaattag

五 (2) (8)

avipilviacdrstvrrtldkllhyrpsaelfpiivsqdcgheetaqaiasygsavthirqpdlssiavppdhrkfqgyykiarhyrwal gqvfrqfrfpaavvveddlevapdffeyfratypllkadpslwcvsawndngkeqmvdasrpellyrtdffpglgwlllaelwael epkwpkafwddwmrrpeqrqgracirpeisrtmtf grkgvshgqff dqhlkfikinqqfvhftqldlsylqreaydrdflarvyg ${
m apqlqvekvrt}$ ndrkel ${
m gevrvqyt}$ grdsf ${
m kafakalgvmddlksgvpragyrgivtfqfpgrrvhlappptwegydpswn}^*$

GnT1 Cys121Thr

agetettececcateategttagecaggaetgegggeacgaggagagaeggeceaggecategeetectaeggeagegggggeagegeae gecetgagateteaagaacgatgaectttggeegeaagggtgtgageeaegggeagttetttgaeeageaeeteaagtttateaagetg aaccagcagtttgtgcacttcacccagctggacctgtcttacctgcagcgggaggcctatgaccgagatttcctcgcccgcgtctacgg cagetteaaggetttegecaaggetetgggtgteatggatgaeettaagteggggggtteegagagetggetaeegggggtattgteaeett getgggegetgggecaggtetteeggeagtttegetteeeegeggeegtggtggtggaggatgaeetggaggtggeeeeggaettet tegagtaetttegggecacetateegetgetgaaggeegaceeteetgtggtgegteteggeetggaatgaeaaeggeaaggage ${ t Gcggtgattcccatcctggtcatcgcctgtgaccgcagcactgttcggcgca{ t ctagacaagctgctgcattatcggccctcggctg}$ atccggcagcccgacctgagcagcattgcggtgccgccggaccaccgcaagttccaggggctactacaagatcgcgcgcactacc ccagttcccgggccgccgtgtccacctggcgcccccaccgacgtgggagggctatgatcctagctggaattag

FIG. 0

lgqvfrqfrfpaavvveddlevapdffeyfratypllkadpslwcvsawndngkeq mvdasrpellyrtdffpglgwlllaelwaendgraffpglgwllaelwaendgraffpglgwllaelwaendgraffpglgwllaelwaendgraffpglgwllaelwaendgraffpglgwllaelwaendgraffpglgwllaelwaendgraffpglgwllaelwaendgraffpglgwllaelwaendgraffglgwllaavipilviacdrstvrraldkllhyrpsaelfpiivsqdcgheetaqaiasygsavthirqpdlssiavppdhrkfqgyykiarhyrwa lepkwpk af wddwmrrpeqrqgracirpe is rtmtfgrkgvshgqffdqhlkfik Inqqfvhftqldlsylqreaydrdflarvyg ${
m apqlqvekvrtndrkelgevrvqytgrdsfkafakalgvmddlksgvpragyrgivtfqfpgrrvhlappptwegydpswn^*}$

GnT1 Cys121Ala

cagettcaaggetttegecaaggetetgggtgtcatggatgacettaagteggggggtteegagagetggetaeeggggtattgteaeett gecetgagateteaagaacgatgacetttggeegeaagggtgtgageeaegggeagttetttgaceageaceteaagtttateaagetg aaccagcagtttgtgcacttcacccagctggacctgtcttacctgcagcgggaggcctatgaccgagatttcctcgccgcgtctacgg ${\sf Gcggtgattcccatcctggtcatcgcctgtgaccgcagcactgttcggcgc{\sf gccctagacaagctgctgcattatcggccctcggctg}$ getgggegetgggecaggtettecggcagtttegetteceegeggeegtggtggtggtggaggatgaeetggaggtggeeeeggaettet tegagtactttegggecacetateegetgetgaaggeegaceeteetegtggtgegteteggeetggaatgaeaaeggeaaggage atccggcagcccgacctgagcagcattgcggtgccgccggaccaccgcaagttccagggctactacaagatcgcgcgccactacc ccagttcccgggccgccgtgtccacctggcgcccccaccgacgtgggaggggtatgatcctagctggaattag

FIG. 10

algqv frqfrfp a avvveddlev apd ffey fratypllk adpslwcvs awn dngkeq mvd as rpellyrtd ffpglgwlllaelwaard and frather appears to be a simple of the first of the f $avipilvia c drstvr{\bf a}{\bf h} ldkllhyrps a elfpiivs qdcgheet aqaiasygs avthirqpdls siavppdhrkfqgyykiar hyrwanipilvia c drstvr{\bf a}{\bf h} ldkllhyrps a elfpiivs qdcgheet aqaiasygs avthirqpdls siavppdhrkfqgyykiar hyrwanipilvia c drstvr{\bf a}{\bf h} ldkllhyrps a elfpiivs qdcgheet aqaiasygs avthirqpdls siavppdhrkfqgyykiar hyrwanipilvia c drstvr{\bf a}{\bf b} ldkllhyrps a elfpiivs qdcgheet aqaiasygs avthirqpdls siavppdhrkfqgyykiar hyrwanipilvia c drstvr{\bf a}{\bf b} ldkllhyrps a elfpiivs a elfpi$ elepkwpk af wddwmrrpeqrqgrac irpe is rtmtfgrkgvshgqffdqhlkfiklnqqfvhftqldlsylqreaydrdflar vyndelepkwpk af weden af the standard of the stand $gapqlqvekvrtudrkelgevrvqytgrdsfkafakalgvmddlksgvpragyrgivtfqfpgrrvhlappptwegydpswn^{\ast}$

GnT1 Arg120Ala, Cys121H

gocotgagatotoaagaacgatgacotttggccgcaagggtgtgagccacgggcagttctttgaccagcacctcaagttlatcaagctg cagettcaaggetttegecaaggetetgggtgtcatggatgacettaagteggggggtteegagagetggetaeeggggtattgteaeett $\operatorname{igctcccagctgcaggtggagaaagtgaggaccaatgaccggaaggagctgggggaggtgcgggtgcagtatacggggcaggag}$ aaccagcagtttgtgcacttcacccagctggacctgtcttacctgcagcgggaggcctatgaccgagatttcctcgcccgcgtctacgg ${\sf Gegggggatteccatectggtcatcgcctgtgaccgcagcactgttcgg} {\sf geceac}$ gctgggcgctgggccaggtcttccggcagtttcgcttccccgcgggccgtggtggtggaggatgacctggaggtggccccggacttct ccagttcccgggccgccgtgtccacctggcgcccccaccgacgtgggaggggggtatgatcctagctggaattag

EIG. 11

Rat Liver ST3Gal III amino acid sequence:

ARIREFVPPFGIKGQDNLIKAILSVTKEYRLTPALDSLHCRRCIIVGNGGVLANKSLGS RIDDYDIVIRLNSAPVKGFEKDVGSKTTLRITYPEGAMQRPEQYERDSLFVLAGFKW <u>MGLLVFVRNLLLALCLFLVLGFLYYSAW</u>KLHLLQWEDSNSLILSLDSAGQTLGTEYDRL GFLLKLDSKLPAELATKYANFSEGACKPGYASAMMTAIFPRFSKPAPMFLDDSFRKW GLMGRGNIPTLGSVAVTMALDGCDEVAVAGFGYDMNTPNAPLHYYETVRMAAIKE QDFKWLKYIVYKERVSASDGFWKSVATRVPKEPPEIRILNPYFIQEAAFTLIGLPFNN SWTHNIQREKEFLRKLVKARVITDLSSGI

FIG. 12

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Full length UDP-N-acetylgalactosaminyltransferase 2 (GalNAcT2)
nucleic acid and amino acid sequences

Amino acid sequence Met Arg Arg Ser Arg Met Leu Leu Cys Phe Ala Phe Leu Trp Val 1 5 15 Leu Gly Ile Ala Tyr Tyr Met Tyr Ser Gly Gly Gly Ser Ala Leu Ala 20 25 30 Gly Gly Ala Gly Gly Ala Gly Arg Lys Glu Asp Trp Asn Glu Ile \cdot 35 40 45 Asp Pro Ile Lys Lys Lys Asp Leu His His Ser Asn Gly Glu Glu Lys 50 55 Ala Gln Ser Met Glu Thr Leu Pro Pro Gly Lys Val Arg Trp Pro Asp 65 70 75 Phe Asn Glu Ala Tyr Val Gly Gly Thr Met Val Arg Ser Gly Gln Asp Pro Tyr Ala Arg Asn Lys Phe Asn Gln Val Glu Ser Asp Lys Leu 105 110 105 Arg Met Asp Arg Ala Ile Pro Asp Thr Arg His Asp Gln Cys Gln Arg 115 120 125 Lys Gln Trp Arg Val Asp Leu Pro Ala Thr Ser Val Val Ile Thr Phe 140 His Asn Glu Ala Arg Ser Ala Leu Leu Arg Thr Val Val Ser Val Leu 155 Lys Lys Ser Pro Pro His Leu Ile Lys Glu Ile Ile Leu Val Asp Asp 175 Tyr Ser Asn Asp Pro Glu Asp Gly Ala Leu Leu Gly Lys Ile Glu Lys 180 190 Val Arg Val Leu Arg Asn Asp Arg Arg Glu Gly Leu Met Arg Ser Arg 195 200 205 Val Arg Gly Ala Asp Ala Ala Gln Ala Lys Val Leu Thr Phe Leu Asp 210 215 220 Ser His Cys Glu Cys Asn Glu His Trp Leu Glu Pro Leu Glu Arg 235 Val Ala Glu Asp Arg Thr Arg Val Val Ser Pro Ile Ile Asp Val Ile 245 250 255 Asn Met Asp Asn Phe Gln Tyr Val Gly Ala Ser Ala Asp Leu Lys Gly 260 265 270 Gly Phe Asp Trp Asn Leu Val Phe Lys Trp Asp Tyr Met Thr Pro Glu 280 285

FIG. 13A SUBSTITUTE SHEET (RULE 26)

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Gln Arg Arg Ser Arg Gln Gly Asn Pro Val Ala Pro Ile Lys Thr Pro 295 300 Met Ile Ala Gly Gly Leu Phe Val Met Asp Lys Phe Tyr Phe Glu Glu 310 Leu Gly Lys Tyr Asp Met Met Met Asp Val Trp Gly Glu Asn Leu 325 Glu Ile Ser Phe Arg Val Trp Gln Cys Gly Gly Ser Leu Glu Ile Ile Pro Cys Ser Arg Val Gly His Val Phe Arg Lys Gln His Pro Tyr Thr 365 360 Phe Pro Gly Gly Ser Gly Thr Val Phe Ala Arg Asn Thr Arg Arg Ala 370 380 Ala Glu Val Trp Met Asp Glu Tyr Lys Asn Phe Tyr Tyr Ala Ala Val 395 Pro Ser Ala Arg Asn Val Pro Tyr Gly Asn Ile Gln Ser Arg Leu Glu Leu Arg Lys Leu Ser Cys Lys Pro Phe Lys Trp Tyr Leu Glu Asn 425 Val Tyr Pro Glu Leu Arg Val Pro Asp His Gln Asp Ile Ala Phe Gly 440 Ala Leu Gln Gln Gly Thr Asn Cys Leu Asp Thr Leu Gly His Phe Ala Asp Gly Val Val Gly Val Tyr Glu Cys His Asn Ala Gly Gly Asn Gln 475 465 Glu Trp Ala Leu Thr Lys Glu Lys Ser Val Lys His Met Asp Leu Cys 490 Leu Thr Val Val Asp Arg Ala Pro Gly Ser Leu Ile Lys Leu Gln Gly Cys Arg Glu Asn Asp Ser Arg Gln Lys Trp Glu Gln Ile Glu Gly Asn 515 Ser Lys Leu Arg His Val Gly Ser Asn Leu Cys Leu Asp Ser Arg Thr 540 Ala Lys Ser Gly Gly Leu Ser Val Glu Val Cys Gly Pro Ala Leu Ser 550 Gln Gln Trp Lys Phe Thr Leu Asn Leu Gln Gln 570

FIG. 13A (CONT.) SUBSTITUTE SHEET (RULE 26)

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Nucleic acid sequence atgaggage getagetetge ttagaettae tgtaggtaet aggeatagae	60
tactacatgt actogogogo cogoctotogo otogocogogo ocogogogogo	120
aggaaggagg actggaatga aattgaccc attaaaaaga aagaccttca tcacagcaat	180
ggagaagaga aagcacaaag catggagacc ctccctccag ggaaagtacg gtggccagac	240
tttaaccagg aagcttatgt tggagggacg atggtoogct cogggcagga cocttacgcc	300
ogcaacaagt tcaaccaggt ggagagtgat aagettogaa tggacagage catcoctgae	360
accoggicatig accagitigtica goggiaagicag tiggogggtigg atctgooggic caccagogitig	420
gtgatcacgt ttcacaatga agocaggtog gooctactca ggacogtggt cagogtgctt	480
aagaaaagoc ogooccatct cataaaagaa atcatcttgg tggatgacta cagcaatgat	540
octgaggaog gggototott ggggaaaatt gagaaagtgo gagttottag aaatgatoga	600
agagaaggac toatgagete aagggttagg ggggaagatg etgaaxaage caaggtaetg	660
acettectgg acagteactg cgagtgtaat gagcactggc tggagccct cetggaaagg	720
gtggggggg acaggactog ggttgtgtca cocatcatog atgtcattaa tatggacaac	780
tttcagtatg tgggggcatc tgctgacttg aagggcggtt ttgattggaa cttggtattc	840
aagtgggatt acatgaogoc tgagcagaga aggtoooggc aggggaacoc agtogoocot	900
ataaaaaccc ccatgattgc tggtgggctg tttgtgatgg ataagttcta ttttgaagaa	960
ctggggaagt acgacatgat gatggatgtg tggggaggag agaacctaga gatctcgttc	1020
agagtgtggc agtgtggtgg cagactggag atcatocogt gcagcogtgt gggacacgtg	1080
ttooggaagc agcacoccta cacgittocog ggtggcagtg gcactgtett tgccogaaac	1140
accognoggy cagcagaggt ctggatggat gaatacaaaa atttctatta tgcagcagtg	1200
octtotgota gaaaogttoo ttatggaaat attoagagoa gattggagot taggaagaaa	1260
ctcagctgca agoctttcaa atggtacctt gaaaatgtct atccagagtt aagggttcca	1320
gaccatcagg atatagettt tggggeettg cagcagggaa ctaactgeet cgacaetttg	1380
ggacactttg ctgatggtgt ggttggagtt tatgaatgtc acaatgctgg gggaaaccag	1440
gaatgggoot tgaogaagga gaagtoggtg aagcacatgg atttgtgoot tactgtggtg	1500
gaccoggicac oggictatat tataaagotig caggictigoo gagaaaatiga cagcagacag	1560
aaatgggaac agatogaggg caactocaag ctgaggcacg tgggcagcaa cctgtgcctg	1620
gacagtogca oggoczagag ogggggocta agogtogagg tototogocc ggocctttog	1680
cagcagtgga agttcacgct caacctgcag cag	1713

FIG. 13B SUBSTITUTE SHEET (RULE 26)

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16/54 Δ51 UDP-N-acetylgalactosaminyltransferase 2, GalNAcT2, nucleic acid and amino acid sequences

Amino acid sequence Lys Lys Lys Asp Leu His His Ser Asn Gly Glu Glu Lys Ala Gln Ser 1 10 15 Met Glu Thr Leu Pro Pro Gly Lys Val Arg Trp Pro Asp Phe Asn Gln 20 25 Glu Ala Tyr Val Gly Gly Thr Met Val Arg Ser Gly Gln Asp Pro Tyr 35 40 45 Ala Arg Asn Lys Phe Asn Gln Val Glu Ser Asp Lys Leu Arg Met Asp 50 60 Arg Ala Ile Pro Asp Thr Arg His Asp Gln Cys Gln Arg Lys Gln Trp 65 75 80 Arg Val Asp Leu Pro Ala Thr Ser Val Val Ile Thr Phe His Asn Glu 85 90 95 Ala Arg Ser Ala Leu Leu Arg Thr Val Val Ser Val Leu Lys Lys Ser Pro Pro His Leu Ile Lys Glu Ile Ile Leu Val Asp Asp Tyr Ser Asn 115 120 125 Asp Pro Glu Asp Gly Ala Leu Leu Gly Lys Ile Glu Lys Val Arg Val 130 135 140 Leu Arg Asn Asp Arg Arg Glu Gly Leu Met Arg Ser Arg Val Arg Gly 145 150 160 Ala Asp Ala Ala Gln Ala Lys Val Leu Thr Phe Leu Asp Ser His Cys 165 Glu Cys Asn Glu His Trp Leu Glu Pro Leu Leu Glu Arg Val Ala Glu 180 185 190 Asp Arg Thr Arg Val Val Ser Pro Ile Ile Asp Val Ile Asn Met Asp 195 200 Asn Phe Gln Tyr Val Gly Ala Ser Ala Asp Leu Lys Gly Gly Phe Asp 210 220 Trp Asn Leu Val Phe Lys Trp Asp Tyr Met Thr Pro Glu Gln Arg Arg 225 230 230 Ser Arg Gln Gly Asn Pro Val Ala Pro Ile Lys Thr Pro Met Ile Ala 250 255 Gly Gly Leu Phe Val Met Asp Lys Phe Tyr Phe Glu Glu Leu Gly Lys 260 265 Tyr Asp Met Met Met Asp Val Trp Gly Gly Glu Asn Leu Glu Ile Ser 275 280 285 Phe Arg Val Trp Gln Cys Gly Gly Ser Leu Glu Ile Ile Pro Cys Ser

FIG. 14A SUBSTITUTE SHEET (RULE 26)

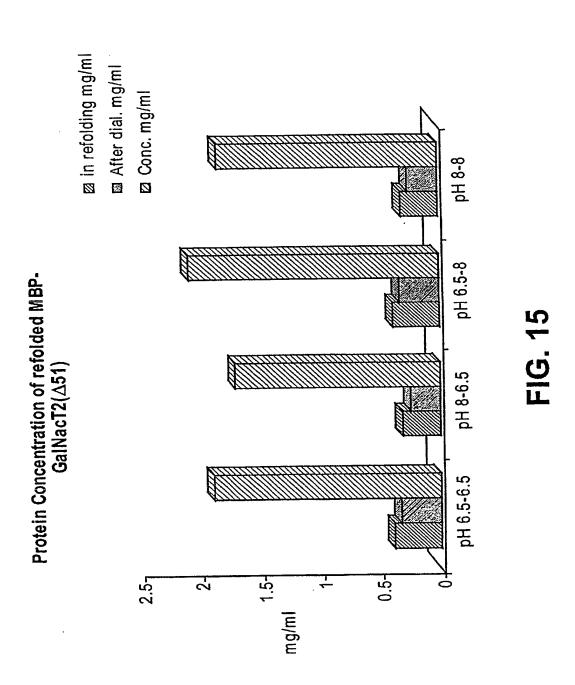
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Arg 305	Val	Gly	His	Val	Phe 310	Arg	Lys	Gln	His	Pro 315	Tyr	Thr	Phe	Pro	Gly 320
Gly	Ser	Gly	Thr	Val 325	Phe	Ala	Arg	Asn	Thr 330	Arg	Arg	Ala	Ala	Glu 335	Val
Trp	Met	Asp	Glu 340	Tyr	Lys	Asn	Phe	Tyr 345	Tyr	Ala	Ala	Val	Pro 350	Ser	Ala
Arg	Asn	Val 355	Pro	Tyr	Gly	Asn	Ile 360	Gln	Ser	Arg	Leu	Glu 365	Leu	Arg	Lys
Lys	Leu 370	Ser	Cys	Lys	Pro	Phe 3 7 5	Lys	Trp	Tyr	Leu	Glu 380	Asn	Val	Tyr	Pro
Glu 385	Leu	Arg	Val	Pro	Asp 390	His	Gln	Asp	Ile	Ala 395	Phe	Gly	Ala	Leu	Gln 400
Gln	Gly	Thr	Asn	Cys 405	Leu	Asp	Thr	Leu	Gly 410	His	Phe	Ala	Asp	Gly 415	Val
Val	Gly	Val	Tyr 420	Glu	Cys	His	Asn	Ala 425	Gly	Gly	Asn	Gln	Glu 430	Trp	Ala
Leu	Thr	Lys 435	Glu	Lys	Ser	Val	Lys 440	His	Met	Asp	Leu	Cys 445	Leu	Thr	Val
Val	Asp 450		Ala	Pro	Gly	Ser 455	Leu	Ile	Lys	Leu	Gln 460	Gly	Cys	Arg	Glu
Asn 465		Ser	Arg	Gln	Lys 470	Trp	Glu	Gln	Ile	Glu 475	Gly	Asn	Ser	Lys	Leu 480
Arg	His	Val	Gly	Ser 485	Asn	Leu	Cys	Leu	Asp 490	Ser	Arg	Thr	Ala	Lys 495	Ser
Gly	Gly	Leu	Ser 500		Glu	Val	Cys	Gly 505	Pro	Ala	Leu	Ser	Gln 510	Gln	Trp
Lys	Phe	Thr 515		Asn	Leu	Gln	Gln 520								

FIG. 14A (CONT.)

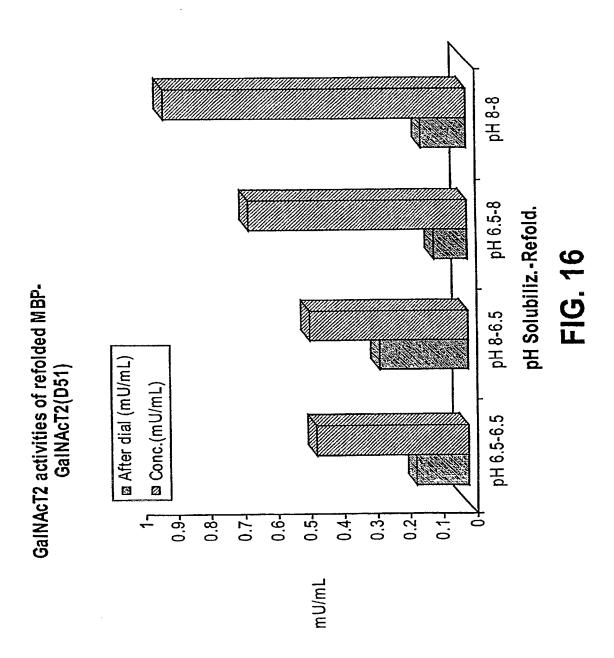
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ctoraggia aajtacijte gezajactt aacagjaaj ettatijtig agjjacjatij 120 gleogeteng gezajace teacjone aacaajtika acaajtiga gajtataag 180 ettogatig acagogoat eerigacae egjeatjae ajtijezgog gaageajtij 240 egjitgjate teorijoorae eagotigija ateacitte acaatgaage eegjitgjac 300 etaeteajja engligitaaj eigtettaaj aaaageme eerateetat aaaagaaate 360 atettogitgi atgaetacaj eaatgateet gaagaegiji eteetetigij gaaaattigag 420 aaagtgegij teettagaa tijatogaaj gaagjetea tijoogetaaj gitaaatgag 540 eaetigetigi agoeeteet gaaagjiji gegjaagjaa gaeetigija gitaaatgag 540 eaetigetigi agoeeteet gaaagjiji gegjaagjaa gaeetigija gijataatgag 660 ateatogatij teattaatat gijaeaaetti eagtatigii gijaateete tijoogetea geagajaagi 720 eteengeaji gijaaeeti gijaatteeaj tijoogatea gaaagaagi 720 eteengeaji gijaaeeti eteenjii gijaattaea tijoogeteja geagajaagi 720 eteengeaji gijaaeeti eteenjii gijaattaea tijoogeteja geagajaagi 720 eteengeaji gijaaeeti eteenjii gijaattaea tijoogeteja geagajaagi 720 eteengeaji gijaaeeti eteenjii gijaagtae aaaaeee eteenjii gijaattaja 1900 ejaajagaaja aeetagaat eteenjii gaagajaaga aaatgatgat gijaatgaat eetijaagat 900 ejaajagaaja aeetagaat eteenjii eegjaajaaga aeetagaata eegiteetij 900 ejaajagaaja eegajataaj gijateeaja eegajajaa eegajajaajaa 1020 eagajagaata eegajataaj gijateeaja eegajajaa eegajajaa eegajajaa 1020 eagajajaata aetijaajaa gajateeaja eegajajaa eegajajaa eegajajaa 1200 eagajajaata aetijaajaa eegajajaa eegajajaa eegajajaa eegajajaa eegajajaa 1200 eagajajaata aetijaajaa eegajajaa e	aaaaagaaag aocttcatca cagcaatgga gaagagaaag cacaaagcat ggagacoctc	60
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cractcaga cogtograca cagostograg atcacottte acaatgaage cagostograc 360 ctactcaga cogtograca cagostograg cagostograc cocatectat acaasgaaate 360 atcttograg atgactacag caatgateet gaggacoggg etectetagg gaaaattaga 420 caagostograg etettagaaa togategaca gaagoetea togacteag gyttogggg 480 cactograg gyactegac etectograca gyactegag gyttogggg 540 cactograg agoocteet gyacaagoggg gyacategag 540 cactograg agoocteet gyacaagoggg gyacategag gyacategag 660 atcategag gyacategag acategagagag acategagaga acategagaga acategagaga acategagaga acategagaga acategagagagagagagagagagagagagagagagagagag		240
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tocogracy gracocagt ogooctata aaaacocca tgattgetgg tgggetgttt 780 gtgatggata agttetattt tgaagaactg gggaagtacg acatgatgat ggatgtgtgg 840 ggaggagga acetagagat etcgttcoge gtgtggcagt gtggtggcag cetggagate 900 atcocgtgca gccgtgtggg acacgtgtte oggaagcage accetacae gttcocgggt 960 ggcagtggca etgtetttge ocgaaacace ogcogggag cagaggtetg gatggatgaa 1020 tacaaaaatt tetattatge agcagtgeet tetgetagaa acgtteetta tggaaatatt 1080 cagagcagat tggagettag gatgaace accettgaa acgtteetta tggaaatatt 1040 aatgtetate cagagttaag ggttcocagae catcaggata tagettttgg ggcettgcag 1200 cagaggacta actgetoga cactttggga cactttgetg atggtgtggt tggagtttat 1260 gaatgteaca atgetgggg aaaaccaggaa tgggeettga ogaggacga gtoggtgaag 1320 cacatggatt tgtgeettae tgtggtggae ogggeacogg getetettat aaagetgcag 1380 ggetgooga aaaatgacag cagacagaaa tgggacagg ocaagaggg gggeetaage 1500 aggcacgtgg gcagcaacet gtgcetggae agtcgcacgg ocaagaggg oggcetaage 1500	atcatogatg toattaatat ggacaacttt cagtatgtgg gggcatctgc tgacttgaag	660
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cacatggatt tgtgccttac tgtggtggac ogggcacogg gctctcttat aaagctgcag 1380 ggctgcogag aaaatgacag cagacagaaa tgggaacaga tcgagggcaa ctccaagctg 1440 aggcacgtgg gcagcaacct gtgcctggac agtcgcacgg ccaagagcgg gggcctaagc 1500	gaatgtcaca atgctggggg aaaccaggaa tgggccttga cgaaggagaa gtcggtgaag	1320
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aggraogtog gragraacet gtgoetggae agtogoacgg coaagagogg gggoetaage 1500		1440
		1500
	gtggaggtgt gtggcoogge cetttegeag eagtggaagt teaegeteaa eetgeageag	1560

FIG. 14B

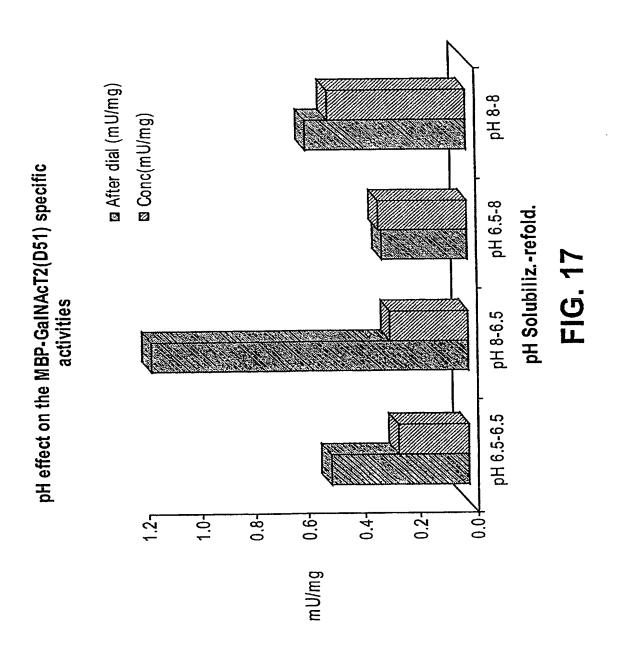
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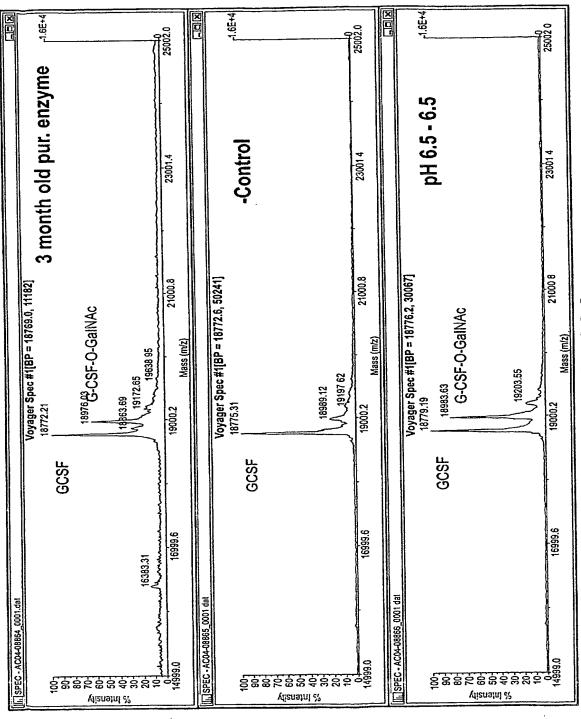


FIG. 18A

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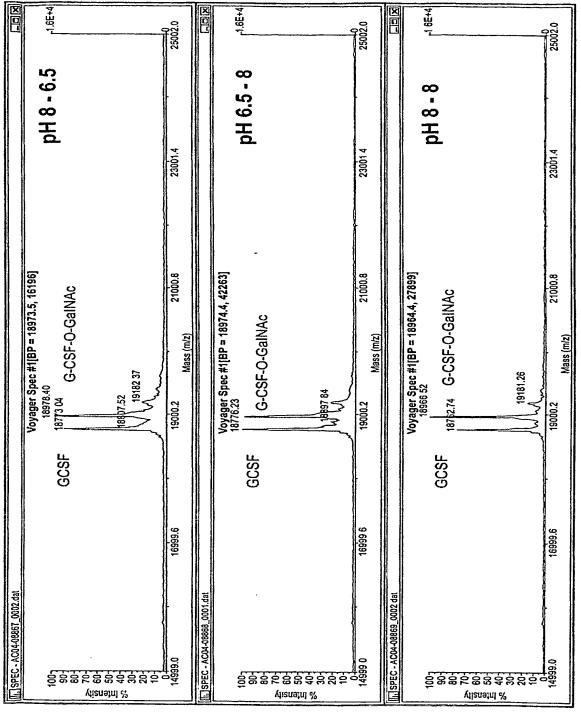
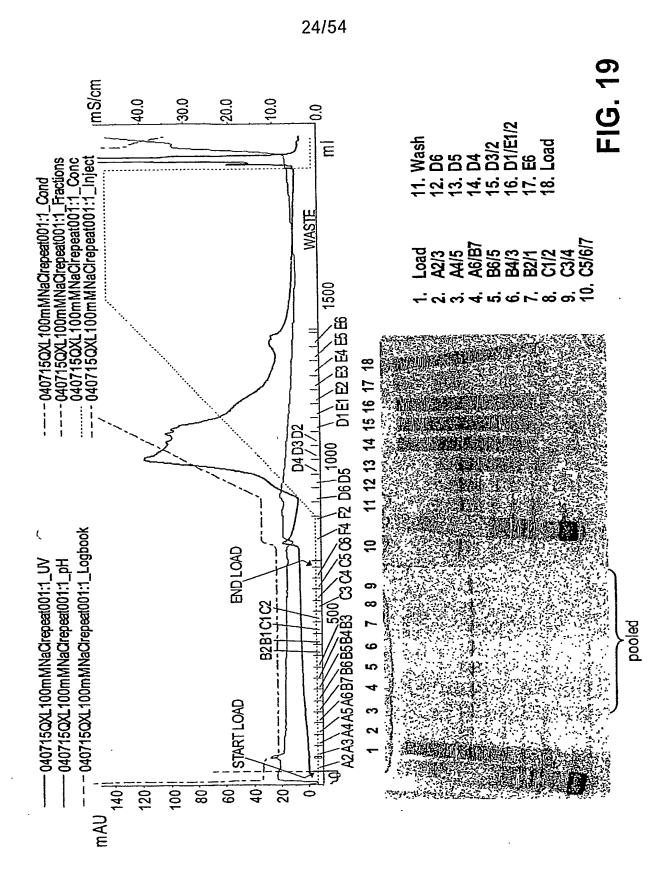


FIG. 18B

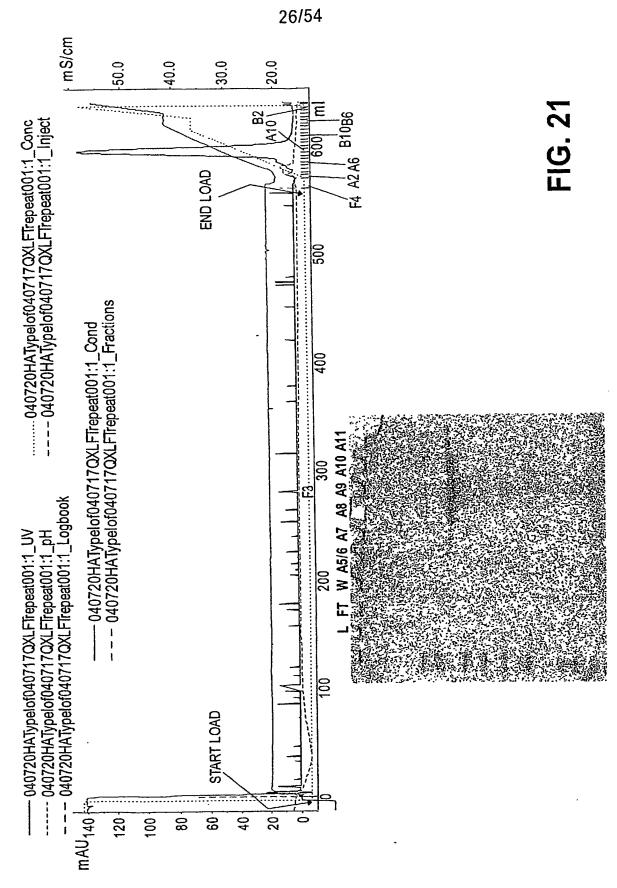


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	Volume (mL)	Activity (U/L)	A280
Load	890	1.5	0.110
FT _{A4-C4}	670	9.2	NA
FT _{C5-C7}	120	1.0	0
Wash	138	3.6	0.100
D6	45	4.5	0
D5	45	2.4	0.026
D4	45	2.0	0.108
D3/2	90	1.1	0.179
E6		0.0	0.017

FIG. 20

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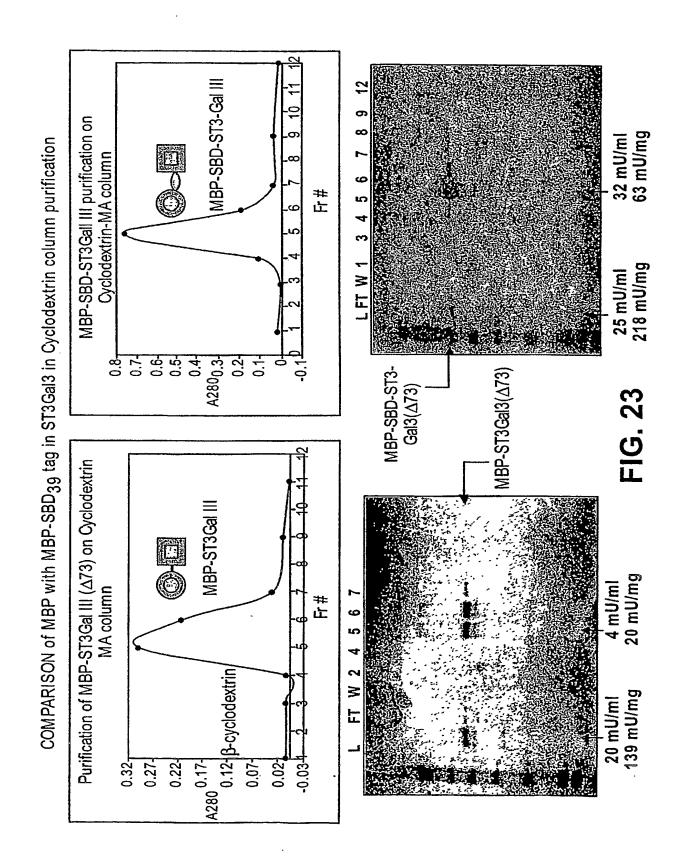
SUBSTITUTE SHEET (RULE 26)

Pooled A4-C4 and adjusted pH to 7.0 using 1mM HCI 670mL load pH 7.0 16mS/cm

	Volume (mL)	Activity (U/L) pre- dialysis	Activity (U/L) post- dialysis	A280	A280/ 1.51 (mg/mL)	Activity (U)	Mass (mg)	Specific Activity (U/mg)
Load	029	9.2	AN	NA				
FT	029	0.0	AN	0.122				
Wash	6	2.9	NA	-0.013				
A5/6	9	1.1	NA	-0.005				
A7	3	0.1						
A8	3	1.3	1					
A9	3	4.6	19.3 (13ml)	0.180	0.119	0.25	1.55	0.16
A10	3	2.4	(10117)					
A11	8	0.4						

FIG. 22

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GNVDPLLEKRLVSCRRCAVVGNSGNLKESYYGPQIDSHDFVLRMNKAPTEGFEADVGSKTTHHFVYPE KELKAKGKSALMFNLQEPYFTWPLIAADGGYAFKYENGKYDIKDVGVDNAGAKAGLTFLVDLIKNKH PCTCTRCIEEORVSAWFDERFNRSMQPLITAKNAHLEEDTYKWWLRLQREKQPNNLNDTIRELFOVVP MNADTDYSIAEAAFNKGETAMTINGPWAWSNIDTSKVNYGVTVLPTFKGQPSKPFVGVLSAGINAASP WYAVRTAVINAASGRQTVDEALKDAQTNSSSNNNNNNNNNNLGIEGRISEFGSELSENFKKLMKYPYR GRYPSTGILSVIFSLHICDEVDLYGFGADSKGNWHHYWENNPSAGAFRKTGVHDGDFESNVTTILASIN NKELAKEFLENYLLTDEGLEAVNKDKPLGAVALKSYEEELAKDPRIAATMENAQKGEIMPNIPQMSAF MKIEEGKLVIWINGDKGYNGLAEVGKKFEKDTGIKVTVEHPDKLEEKFPOVAATGDGPDIIFWAHDRF **SFRELAQEVSMILVPFKTTDLEWVISATTTGRISHT**†VPVPAKIKVKKEKILIYHPAFIKYVFDRWLQGH GGYAQSGLLAEITPDKAFQDKLYPFTWDAVRYNGKLIAYPIAVEALSLIYNKDLLPNPPKTWEEIPALD

MBP-pST3Gal1 fusion protein

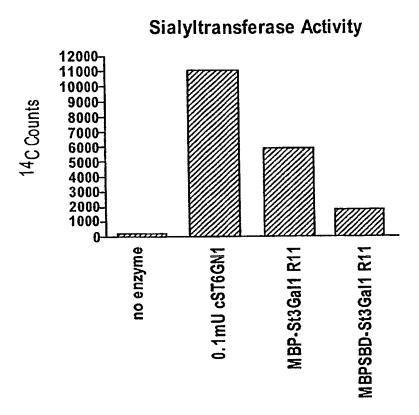
IG. 24A

MBPSBD-pST3Gal1 fusion protein

KELKAKGKSALMFNLQEPYFTWPLIAADGGYAFKYENGKYDIKDVGVDNAGAKAGLTFLVDLIKNKH MINADTDYSIAEAAFNKGETAMTINGPWAWSNIDTSKVNYGVTVLPTFKGQPSKPFVGVLSAGINAASP FKKLMKYPYRPCTCTRCIEEORVSAWFDERFNRSMOPLLTAKNAHLEEDTYKWWLRLOREKOPNNLN DTIRELFQVVPGNVDPLLEKRLVSCRRCAVVGNSGNLKESYYGPQIDSHDFVLRMNKAPTEGFEADVG YVFDRWLQGHGRYPSTGILSVIFSLHICDEVDLYGFGADSKGNWHHYWENNPSAGAFRKTGVHDGDF NKELAKEFLENYLLTDEGLEAVNKDKPLGAVALKSYEEELAKDPRIAATMENAQKGEIMPNIPQMSAF MKIEEGKLVIWINGDKGYNGLAEVGKKFEKDTGIKVTVEHPDKLEEKFPOVAATGDGPDIIFWAHDRF SKTTHHFVYPESFRELAQEVSMILVPFKTTDLEWVISATTTGRISHTYVPVPAKIKVKKEKILIYHPAFIK KYTSSDPLWYVTVTLPAGESFEYKFIRIESDDSVEWESDPNREYTVPQACGTŠTATVTDTWRGSELSEN **GGYAQSGLLAEITPDKAFQDKLYPFTWDAVRYNGKLIAYPIAVEALSLIYNKDLLPNPPKTWEEIPALD** SGSVTSTSKTTATASKTSTSTSSTSCTTPTAVAVTFDLTATTTYGENIYLVGSISQLGDWETSDGIALSAD WYAVRTAVINAASGRQTVDEALKDAQTNSSSNNNNNNNNNNNLGIEGRISEFGSIVATGGTTTTAÌPTG ESNVITILASINKIRIFKGR

FIG. 24B

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Test pST3Gal1 for activity after Hampton refold

FIG. 25

KELKAKGKSALMFNLQEPYFTWPLIAADGGYAFKYENGKYDIKDVGVDNAGAKAGLTFLVDLIKNKH NCSASVKIKASKSPWLQNIFLPNITLFLDSGRFTQSEWNRLEHFAPPFGFMELNQSLVQKVVTRFP HRPQEAFRNALDLDRYLLLHPDFLRYMKNRFLRSKTLDTAXWRIYRPTTGALLLLTALHLCDKV MINADTDYSIAEAAFNKGETAMTINGPWAWSNIDTSKVNYGVTVLPTFKGQPSKPFVGVLSAGINAASP STRKTPMATGAVPAKKKVVQATKSPASSPHPTTRRRQRLKASEFKSEPRWDFEEEYSLDMSSLQT SFYGFTAFSLTQSILILGRRGFQHVPLGKDVRYLHFLEGTRNYEWLEAMFLNQTLAKTHLSWFR PVRQQQLLLASLPTGYSKCITCAVVGNGGILNDSRVGREIDSHDYVFRLSGAVIKGYEQDVGTRT NKELAKEFLENYLLTDEGLEAVNKDKPLGAVALKSYEEELAKDPRIAATMENAQKGEIMPNIPQMSAF WYAVRTAVINAASGRQTVDEALKDAQTNSSSNNNNNNNNNNLGIEGRISEFGSSEHLDKVPRTPGAL GGYAQSGLLAEITPDKAFQDKLYPFTWDAVRYNGKLIAYPIAVEALSLIYNKDLLPNPPKTWEEIPALD SAYGFTTEGHERFSDHYYDTSWKRLIFYINHDFRLERMVWKRLHDEGIIWLYQRPQSDKAKN

MKIEEGKLVIWINGDKGYNGLAEVGKKFEKDTGIKVTVEHPDKLEEKFPQVAATGDGPDIIFWAHDRI

FIG. 26A

GILNNSHIMGQEIDSHDYVFRLSGALIKGYEQDVGTRTSFYGFTAFSLTQSLLILGNRGFKNVPLGK NIKERSLQSLAKPKSQAPTRARRITIYAEPVPENNALNTQTQPKAHTTGDRGKEANQAPPEEQDK SPTTQRNQRLKAANFKSEPRWDFEEKYSFEIGGLQTTCPDSVKIKASKSLWLQKLFLPNLTLFLDS KNRFLRSKTLDGAHWRIYRPTTGALLLLTALQLCDQVSAYGFTTEGHERFSDHYYDTSWKRLJFY KELKAKGKSALMFNLQEPYFTWPLIAADGGYAFKYENGKYDIKDVGVDNAGAKAGLTFLVDLIKNKH DVRYLHFLEGTRDYEWLEALLMNQTVMSKNLFWFRHRPQEAFREALHMDRYLLLHPDFLRYM **VPHTAQRAAWKSPEKEKTMVNTLSPRGQDAGMASGRTEAQSWKSQDTKTTQGNGGQTRKLTA** MNADTDYSIAEAAFNKGETAMTINGPWAWSNIDTSKVNYGVTVLPTFKGQPSKPFVGVLSAGINAASP NKELAKEFLENYLLTDEGLEAVNKDKPLGAVALKSYEEELAKDPRIAATMENAQKGEIMPNIPQMSAF WYAVRTAVINAASGRQTVDEALKDAQTNSSSNNNNNNNNNNLGIEGRISEFGSKEPQTKPSRHQKTE MKIEEGKLVIWINGDKGYNGLAEVGKKFEKDTGIKVTVEHPDKLEEKFPQVAATGDGPDIIFWAHDRF SRIVSEKHQGKAATTAKTLIPKSQHRMLAPTGAVSTRIRQKGVTTAVIPPKEKKPQATPPPAPFQ RHFNQSEWDRLEHFAPPFGFMELNYSLVQKVVTRFPPVPQQQLLLASLPAGSLRCITCAVVGNG **GGYAQSGLLAEITPDKAFQDKLYPFTWDAVRYNGKLIAYPIAVEALSLIYNKDLLPNPPKTWEEIPALD** NHDFKLEREVWKRLHDEGIRLYQRPGPGTAKAKN

FIG. 26E

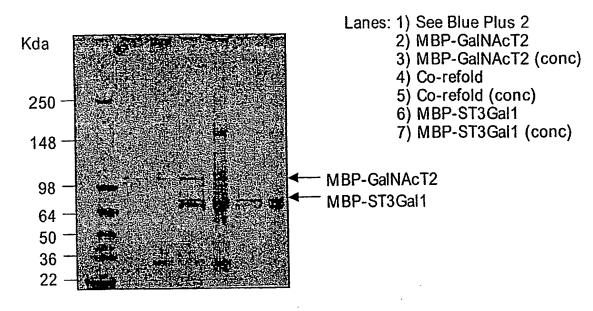


FIG. 27A

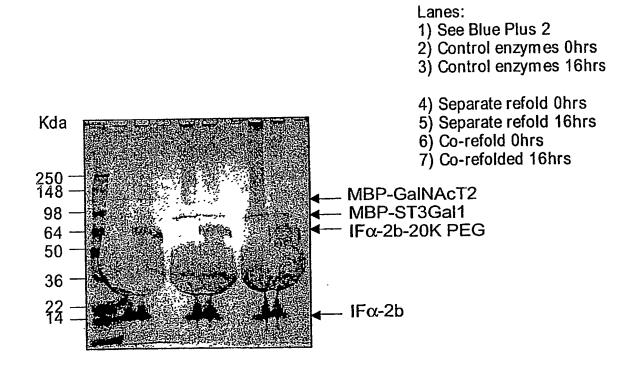
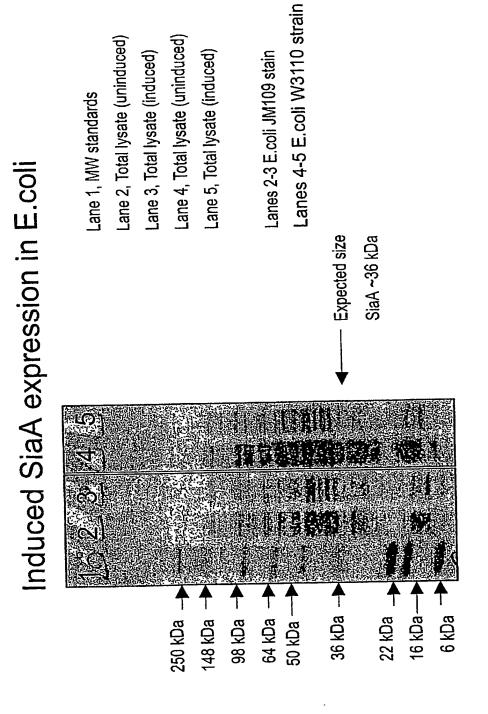


FIG. 27B



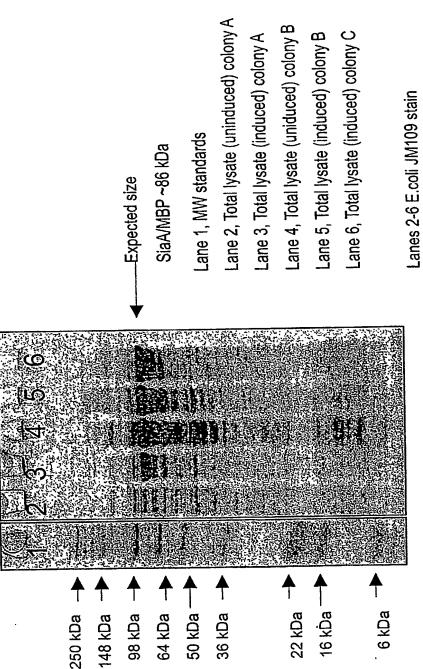
There is no obvious inducible band at the expected

Mass of 36 kDa for the native SiaA protein.

FIG. 28

Induced SiaA/MBP expression in E.coli

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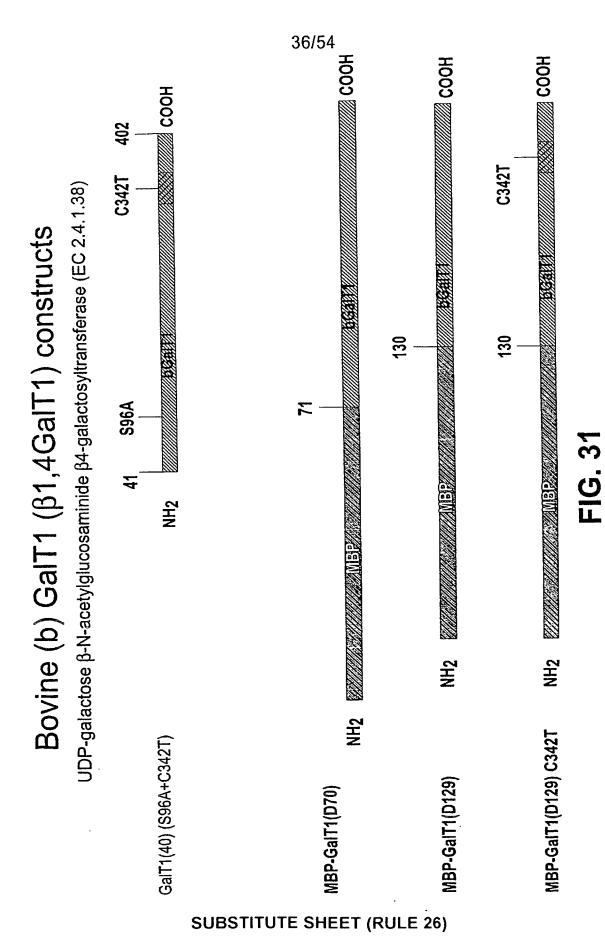


Compare to figure X where SiaA production is not obvious. The presence of High level production of SiaA/MBP even in absence of IPTG induction. the fusion partner (MBP) drives high levels of expression.

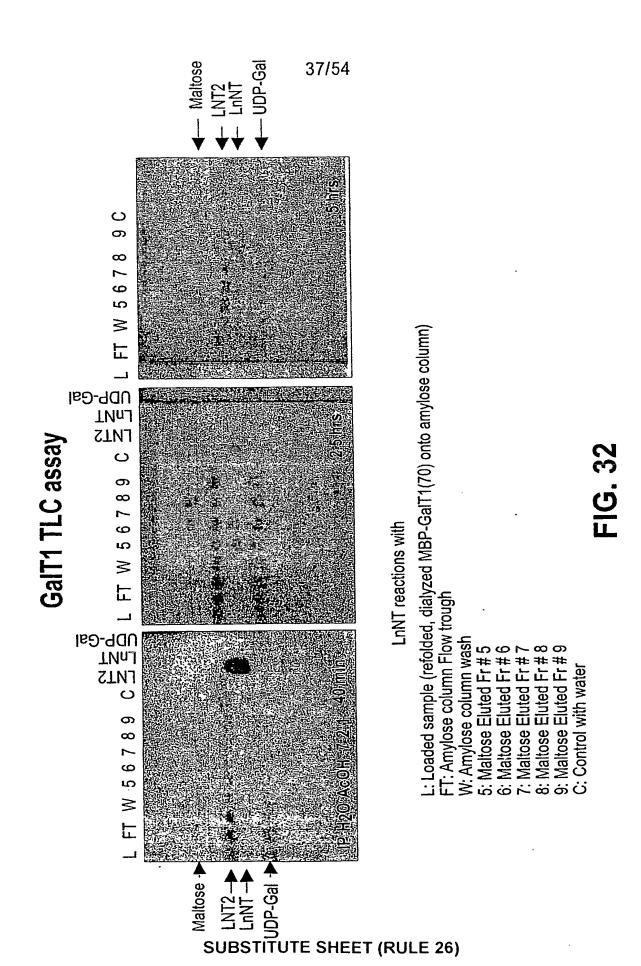
FIG. 29

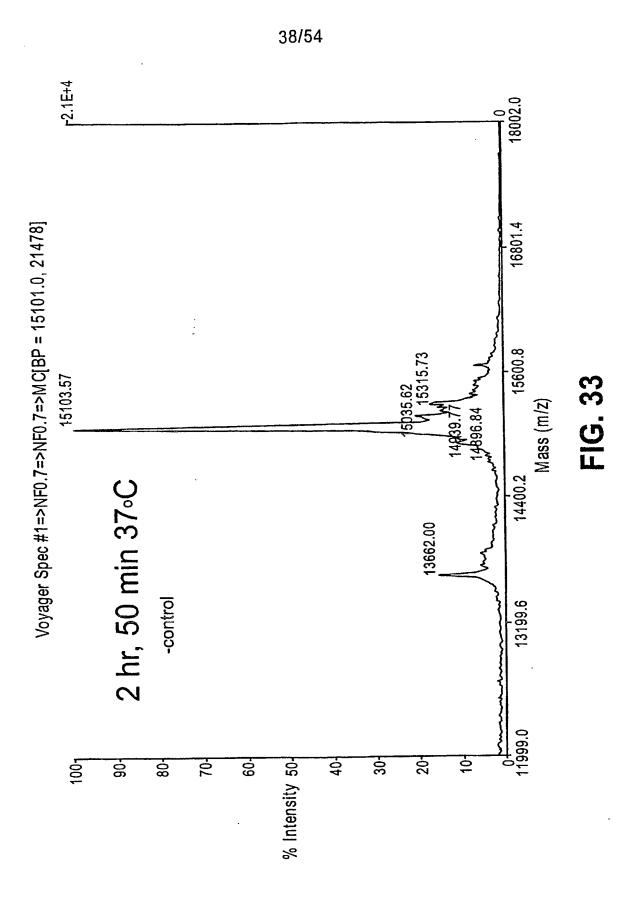
kknepnpgrf gpgpgsnlts ggvsalskqq ytpmdcisph llnvgfkeal pqlvgvhppl alhlgvtlvy ylagrdlrrl agesmfnrak gfslpyvqyf kcrmirhsrd nldayshpgp qnpkvklggr svsrpnavig ytkitvdigt sskprsraps ipvdlklieg ldygiyving rhisvamdkf iynrlafrgm mvlevqrypl lvgpmliefn hntyrcfsqp ylhpmvqrqq gvapppplgn racrllvavc ywgwggeddd lsdglnslty rqehlkywly sdvdlipmnd gqpsgelrlr ltacpeespl saampgaslq kvaiiilfrn flsingfpnn driahtketm kdydyncfvf apvpstttrs mkfrepllgg qgsshgaaai 121 181 241 301

FIG. 3(

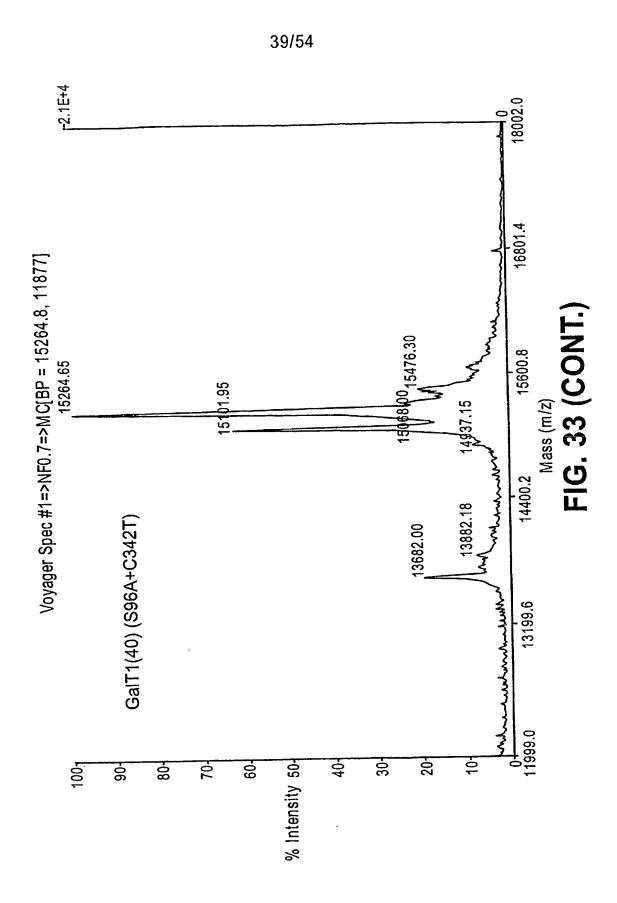


WO 2005/089102 PCT/US2005/003856

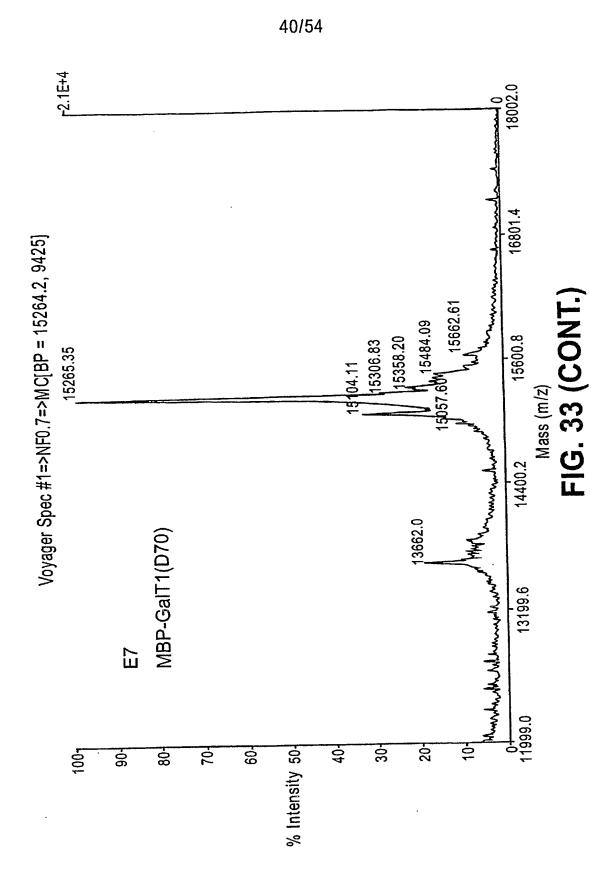


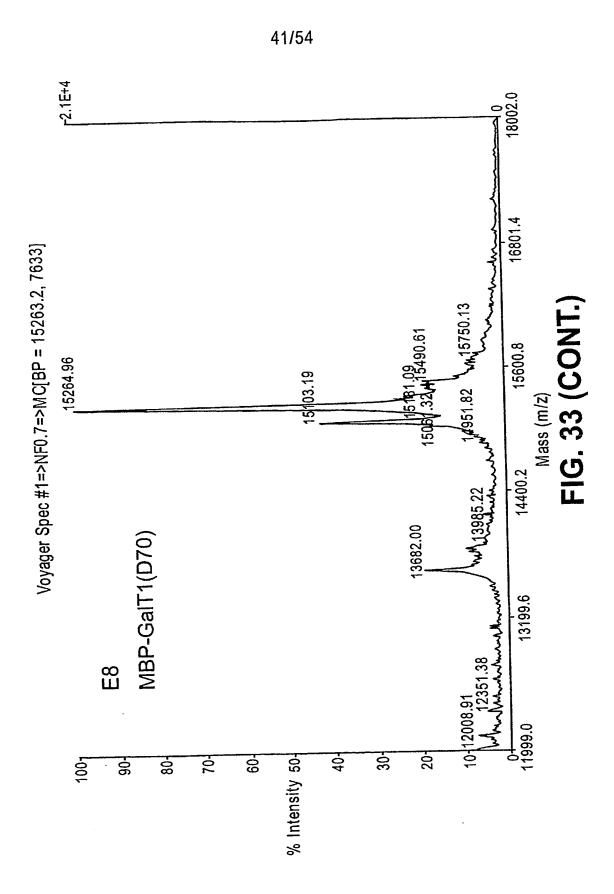


PCT/US2005/003856

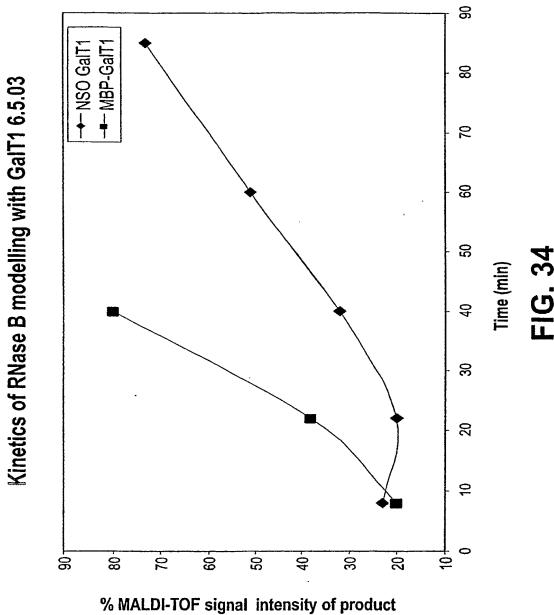


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formation

GnT1 constructs

MBP-GnT1(A35)

MBP-GnT1(Δ103)

mlkkqsaglv lwgailfvaw nallllffwt rpapgrppsv saldgdpasl trevirlagd aevelerqrg llqqigdals sqrgrvptaa ppaqprvpvt papavipilv iacdrstvrr

121 cldkllhyrp saelfpiivs qdcgheetaq aiasygsavt hirqpdlssi avppdhrkfq 181 gyykiarhyr walgqvfrqf rfpaavvved dlevapdffe yfratypllk adpslwcvsa

241 wndngkeqmv dasrpellyr tdffpglgwl llaelwaele pkwpkafwdd wmrrpegrgg

301 racirpeisr tmtfgrkgvs hgqffdqhlk fiklnqqfvh ftqldlsylq reaydrdfla

361 rvygapqlqv ekvrtndrke lgevrvqytg rdsfkafaka lgvmddlksg vpragyrgiv

421 tfqfrgrrvh lappptwegy dpswn

FIG. 35



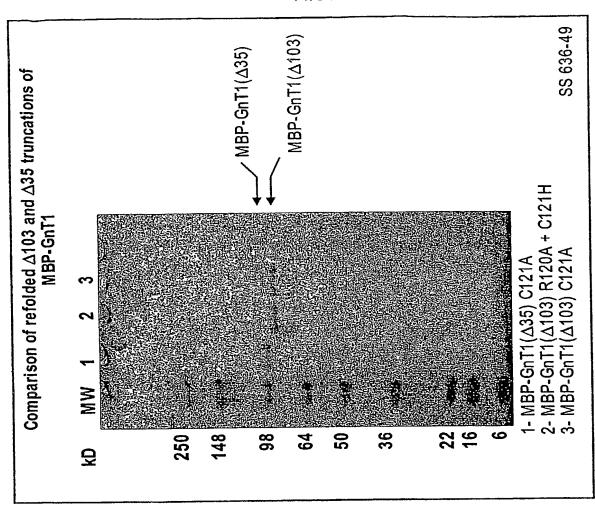
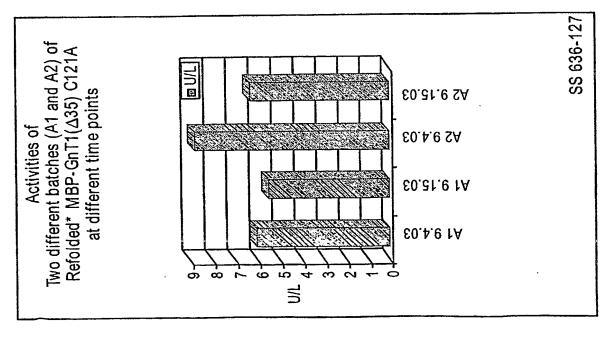


FIG. 36



rmnkaptegf htyvpvpaki gfgadskgnw

isatttgtis

pqidshdfvl

1hicdevdly

stgilsvifs

laqevsmilv rwlqghgryp

gdfesnvtti

hpafikyvfd

kvkkekiliy

241

181

hhywennpsa

gafrktgvhd

hfvypesfre

scrrcavvgn fnrsmqpllt

qrvsawfder

vdpllekrlv

relfqvvpgn eadvgsktth

pctctrciee mapmrkkstl

klltllvlfi

kgpnnlndti fkklmkypyr

ykwwlrlgre

aknahleedt sgnlkesyyg pfkttdlewv

kqmvielsen

fltsfflnys htvvttawfp

Human ST6GalNAcI
MRSCLWRCRHLSQGVQWSLLLAVLVFFLFALPSFIKEPQTKPSRHQRTENIKERSLQS
LAKPKSQAPTRARRTTIYAEPVPENNALNTQTQPKAHTTGDRGKEANQAPPEEQDK
VPHTAQRAAWKSPEKEKTMVNTLSPRGQDAGMASGRTEAQSWKSQDTKTTQGNG
GQTRKLTASRTVSEKHQGKAATTAKTLIPKSQHRMLAPTGAVSTRTRQKGVTTAVIP
PKEKKPQATPPPAPFQSPTTQRNQRLKAANFKSEPRWDFEEKYSFEIGGLQTTCPDSV
KIKASKSLWLQKLFLPNLTLFLDSRHFNQSEWDRLEHFAPPFGFMELNYSLVQKVVT
RFPPVPQQQLLLASLPAGSLRCITCAVVGNGGILNNSHMGQEIDSHDYVFRLSGALIK
GYEQDVGTRTSFYGFTAFSLTQSLLILGNRGFKNVPLGKDVRYLHFLEGTRDYEWLE
ALLMNQTVMSKNLFWFRHRPQEAFREALHMDRYLLLHPDFLRYMKNRFLRSKTLD
GAHWRIYRPTTGALLLLTALQLCDQVSAYGFITEGHERFSDHYYDTSWKRLIFYINH
DFKLEREVWKRLHDEGIIRLYQRPGPGTAKAKN

FIG. 38A

Chicken ST6GalNAcI
MGFLIRRLPKDSRIFRWLLILTVFSFIITSFSALFGMEKSIFRQLKIYQSIAHMLQVDTQ
DQQGSNYSANGRISKVGLERDIAWLELNTAVSTPSGEGKEEQKKTVKPVAKVEEAK
EKVTVKPFPEVMGITNTTASTASVVERTKEKTTARPVPGVGEADGKRTTIALPSMKE
DKEKATVKPSFGMKVAHANSTSKDKPKAEEPPASVKAIRPVTQAATVTEKKKLRAA
DFKTEPQWDFDDEYILDSSSPVSTCSESVRAKAAKSDWLRDLFLPNITLFIDKSYFNV
SEWDRLEHFAPPYGFMELNYSLVEEVMSRLPPNPHQQLLLANSSSNVSTCISCAVVG
NGGILNNSGMGQEIDSHDYVFRVSGAVIKGYEKDVGTKTSFYGFTAYSLVSSLQNLG
HKGFKKIPQGKHIRYIHFLEAVRDYEWLKALLLDKDIRKGFLNYYGRRPRERFDEDF
TMNKYLVAHPDFLRYLKNRFLKSKNLQKPYWRLYRPTTGALLLLTALHLCDRVSAY
GYITEGHQKYSDHYYDKEWKRLVFYVNHDFNLEKQVWKRLHDENIMKLYQRS

FIG. 38B

Mouse ST6GalNAcI protein beginning at residue 32 of the native mouse protein DPRAKDSRCQFIWKNDASAQENQQKAEPQVPIMTLSPRVHNKESTSVSSKDLKKQER EAVQGEQAEGKEKRKLETIRPAPENPQSKAEPAAKTPVSEHLDKLPRTPGALSTRKTP MATGAVPAKKKVVQATKSPASSPHPTTRRRQRLKASEFKSEPRWDFEEEYSLDMSSL QTNCSASVKIKASKSPWLQNIFLPNITLFLDSGRFTQSEWNRLEHFAPPFGFMELNQSL VQKVVTRFPPVRQQQLLLASLPTGYSKCITCAVVGNGGILNDSRVGREIDSHDYVFR LSGAVIKGYEQDVGTRTSFYGFTAFSLTQSILILGRRGFQHVPLGKDVRYLHFLEGTR NYEWLEAMFLNQTLAKTHLSWFRHRPQEAFRNALDLDRYLLLHPDFLRYMKNRFL RSKTLDTAHWRIYRPTTGALLLLTALHLCDKVSAYGFITEGHQRFSDHYYDTSWKRL IFYINHDFRLERMVWKRLHDEGIIWLYQRPQSDKAKN

FIG. 38C

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Hum (h)ST6GalNAcl truncations	N600 S65 aa N600		E110	M134 467 aa	T171 430 aa	73	
	Δ35	Δ72	∆109	Δ133	Δ170	Δ232	Δ272

FIG. 38

MBP-hST6GalNAcl constructs

48/54 MBP-hST6GalNAcl (Δ272), 81.4 kD ☐ MBP-hST6GalNAcl (∆232), 86.0 kD ട്ട**ു** MBP-hST6GalNAcl (∆170), 92.7 kD ☑ MBP-hST6GalNAcl (∆133), 96.7 kD ह्य MBP-hST6GalNAcl (∆109), 99.2 kD ₩ MBP-hST6GalNAcl (Δ72), 103.3 kD MBP-hST6GaINAcl (A35), 107.6 kD ST6 Gain Aci K36

FIG. 40

MASKSWLNFLTFLCGSAIGFLLCSQLFSILLGEKVDTQPNVLHNDPHARHSDDNGQN HLEGQMNFNADSSQHKDENTDIAENLYQKVRILCWVMTGPQNLEKKAKHVKATW AQRCNKVLFMSSEENKDFPAVGLKTKEGRDQLYWKTIKAFQYVHEHYLEDADWFL KADDDTYVILDNLRWLLSKYDPEEPIYFGRRFKPYVKQGYMSGGAGYVLSKEALKR FVDAFKTDKCTHSSSIEDLALGRCMEIMNVEAGDSRDTIGKETFHPFVPEHHLIKGYL PRTFWYWNYNYYPPVEGPGCCSDLAVSFHYVDSTTMYELEYLVYHLRPYGYLYRY QPTLPERILKEISQANKNEDTKVKLGNP

FIG. 41

									50/	54								
			ις C	50	0	100	150	150	000	200	C L	250))	300	300	350	350	
											•							
		C U	50	NSTIAERLYS	100	DDELDAVALP DDELDAVALP	150	VENMRYMLYP VENMRYMLYP	200	EALPNPKLCK FAT DNPKT CK	250	HHLIPSHIDK	300		IYHLRPYGII 350			
		(40	DVGGHEHVHE	90	NKLIFMSSAK NKLIFMSSAK	140	LKADDDTYTI LKADDDTYTI	190	SREAVRREVV	_		RGKFFFF 290		PNOMYVLDYL 340	7777		
Mode	Normal	Normal	30	DMMEMSGPEQ	OMMHMSG-PERO OMMHM	HVKRTWGKRC HVKRTWGKRC	130	EHHINDADWF EHHINDADWF	180		XMSGGAGIVL 230	<u></u>	LAGDSRDSNG 280	DNAI	DNAISFHYVS		EQATESTSDG EQATESTSDG	
		342 N	20	GDVNDAHHSH	GDVNDAHHSH 70	NPSNHOKKAR NPSNHOKKAR	120	KTKEAYKYIY KTKEAYKYIY	170	CKFKPYVKQG	CKFKPYVKQG 220	[IGKCLQNVNV	IGKCLONVNV	KTDEGLDCCS	KIDE	-	AVGELMPEIK AVGELMPEIK	
Range	1-1	1	10	EFMPYDGHRH	EFMPYDGHRH 60	EVRVI.CWIMT EVRVI.CWIMT	110	VGEGRNINLWG	160	YSPETPVYFG	YSPETPVYFG	SDNSGAEDVE	SDNSGAEDVE	Z O O KEWYWOY I FY	KEWYWOYIEY	310	NTPDAL.PNKL NTPDAL.PNKL	
Size	342	342		\leftarrow	H	51	i }	101	TOT	151	151	201	201	251	251		301	i
Sequence	SC1GALT1.AMI	SPTS122.AMI		SC1GALT1.AMI	SPTS122.AMI	SCIGALTI.AMI	SPTSIZZ.AML	SC1GALT1.AMI	SFTST72. AIM	SC1GALT1.AMI	SPTS122.AMI	SC1GALT1.AMI	SPTS122.AMI	TM4 LE #40 100	SPIS122.AMI		SCIGALTI.AMI	

FIG. 42

VAATGDGPDI	WINGDKGYNG IFWAHDRFGG	LAEVGKKFEK YAQSGLLAEL DLVKEAPKTW	FETPALDKTL	RANGKSAIMW
	t 7 T ス ス ひ () () () ス なっ	KRUMGVYDAK	MACAMMAGWA	TODOLLADDA
	~ T T T T T T T T T T T T T T T T T T T	GETAMTINGP SPNKELATEF	1481111006	DWD AMINDINE TO
GAVALKSFOE	QLAKDPRIAA	TMDNATNGEL	MPNIPQMAAF	WYATRSAVLN
ATTGROTVEA	ALNDAATRIT	K		

FIG. 43A

FIG. 43B

Pyrococcus fi MKIEEGKVVI	iriosus MBP		AT ODEWETUE	COKDNITEDAL
MKIEEGKVVÍ	WHAMQPNELE	VEQSLAEEIM	AP25FAFTAE	TMEEDDMAOD
**********	カイ かんりょう ロンかし	12K 6/4 6/4(4) 13 15	PEDBIVIOUS	TIMELTER
	T 17 (7) (1) (1) (1) (1) (1) (1) (1)	TYNKKMMSEP		LIDITAL T DY TYM
	ひつてつみずれへれに	CCVVWIIIKTE	いちにいいひをひてす	FOILT LL TOT
TOTAL PROPERTY	NUMBER	DADMMVNGPW	SINDAVVOR	MEGAATHTT
OTTTTTTDD	DVACUUT TVV	AACIKNKDAA	MVGAVMTITIO	
T CV T PUT TKV	TODPEIKNDP	VIYGFGQAVQ	HAYLMPKSPK	MOGADG
AINEILQDPQ	NADIEGILKK	YQQEILNNMQ	G	

Thermococcus litoralis MBP

MKIEEGKIVF AVGGAPNEIE YWKGVIAEFE KKYPGVTVEL KRQATDTEQR
RLDLVNALRG KSSDPDVFLM DVAWLGQFIA SGWLEPLDDY VQKDNYDLSV
FFQSVINLAD KQGKLYALP VYIDAGLLYY RKDLLEKYGY SKPPETWQEL
VEMAQKIQSG ERETNPNFWG FVWQGKQYEG LVCDFVEYVY SNGGSLGEFK
DGKWVPTLNK PENVEALQFM VDLIHKYKIS PPNTYTEMTE EPVRLMFQQG
NAAFERNWPY AWGLHNADDS SVQKGFAMNL GWNPGRVDVY DDPAVVSKSP
SKYSDNKALA WEFVKFVESY SVQKGFAMNL GWNPGRVDVY DDPAVVSKSP
HLKELRAVFE NAVPRPIVPY YPQLSEIIQK YVNSALAGKI SPQEALDKAQ

FIG. 43D

KEAEELVKQY SK

TAAPQGQGAD IIVG. GGKLYGVPYA MEAV. FIYDVANFYF SAPF RMIDEGVLTP GDNY IPELEPGVPA KPFV ADPRLPARKD VLEL	KOVDIL QKLGEEFRAN AHDWVG ELAVNGLIEP ALIYNK DYVDSVPKTM ILGYGG YVFKETPQGL GTMDSM FKEGLAAMII GVQGFM INAKSPNKVI VKDNPD VVAFTQSASM	DELIEKAKQI DVTDIGLANE NGLWAIKSYK AMEFLTNFIA	DEEYGGEVRG GAVKGAKLIK DAGINYGVAP RKETMYKIYL
ADPRLPARKD VLEL	VKDNPD VVAFTQSASM LKEAVD KIKAQIEK	GIEMENVEEM	ME (MOINIODI)

FIG. 43E SUBSTITUTE SHEET (RULE 26)

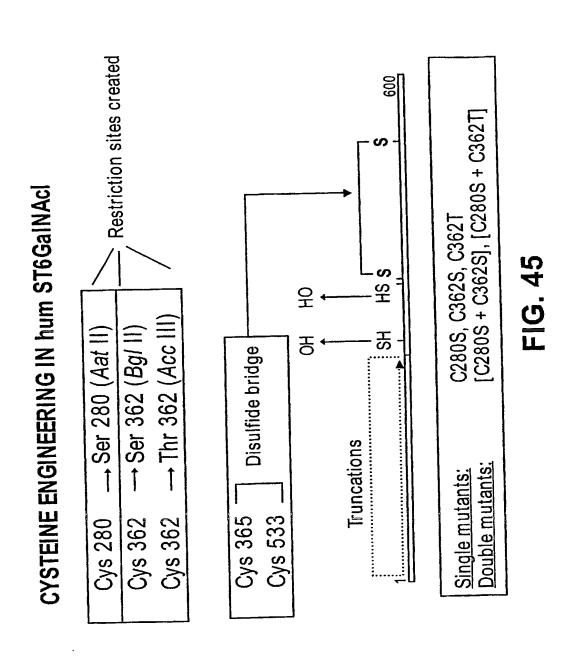
Vibrio choler	ae MBP			DDAT ODKEDO
MKTEEGOLTI	WINGDKGYNG	LAEVGKKFEA	DTGIKVTVAH	POWINGOVERA
AND T	777GCC11 K137711	VARAGLIJVEL	V 52VVTOPG+	V DIIII
	TIPOT OT TVNK	DTMPNPPKSW	FFAVEPDDVV	TITIDOMOTITION
NCKTIGIETH	APOTOTITIES	KACADCADAK	DAGINNKGVK	DAMNFVKGLV
NTKEDAFLMD	LMAADGGIAF	CMMAMMINGD	WSWGNIEKSG	INYGVTTLPK
DKGVISPDMD	YSVSESAFNQ	GNIAMIINGI	TENUT I THING	T.RMVNNDKPL
FNGQASKPFV	GVLTAGISTA	SPNKDLAVEE	IENYLLTNDG	MCCYKNYLLM
GAVALNSFOR	ELDADARIAA	TMDNAMNGEI	WENTEGMNAL	WSSAKNAIIN
TVDGROTVDA	ALADAEKQMT	KP		

FIG. 43F

53/54 1 50 (1)MRKFAYCKVVLATSLIWVLLDMFLLLYFS-----ECNKC HSGALNAT1.pep (1) MRRRS--RMLLCFAFLWVLGIAYYMYSGGGSALAGGAGGGAGRKEDWNEI HSGALNAT2.pep (1)MRK A KMLL A IWVL Consensus FL100 51 (35) DEKKERGLPAGDVLEPVQKPHEGP-G----EMGKPVVIPKEDQEKMKEM HSGALNAT1.pep (49) DPIKKKDLHHSNGEEKAQSMETLPPGKVRWPDFNQEAYVGGIMVRSGQDP HSGALNAT2.pep (51)D KKL D Consensus (79) FKINQFNLMASEMIALNRSLPDVRLEGCKTKVYPDNLPTTSVVIVFHNEA HSGALNAT1.pep (99) YARNKENQVESDKLRMDRAIPDTRHDQCQRKQWRVDLPATSVVITFHNEA HSGALNAT2.pep Consensus (101)F N FN M SD I L RAIPD R D C K W LP TSVVI FHNEA 151 HSGALNAT1.pep (129)WSTLLRTVHSVINRSPRHMIEEIVLVDDASERDFLKRPLESYVKKLKVPV HSGALNAT2.pep (149) RSALLRTVVSVLKKSPPHLIKEIILVDDYSN-----DPEDGALLGKIEKV Consensus (151) S LLRTV SVI KSP HLI EIILVDD S PD L V 250 201 HSGALNATI.pep (179) HVIRMEQRSGLIRARLKGAAVSKGQVITFLDAHCECTVGWLEPLLARIKH HSGALNAT2.pep (194) RVLRNDRREGLMRSRVRGADAAQAKVLTFLDSHCECNEHWLEPLLERVAE Consensus (201) VIR D R GLIRARLKGA A A VITFLDAHCEC WLEPLL RI 251 HSCALNATI.pep (229) DRRIVVCPIIDVISDDTFEYMAGSDMTYGGFNWKLNFRWYPVPQREMDRR HSGAINAT2.pep (244) DRTRVVSPIIDVINMDNFQYVGASADLKGGFDWNLVFKWDYMTPEQRRSR Consensus (251) DR VV PIIDVI D F YMAAS GGF W L FKW M 350 301 HSGALNAT1.pep (279)KGDRTLPVRTPIMAGGLFSIDRDYFQEIGTYDAGMDIWGGENLEISFRIW HSGALNAT2.pep (294)QGNPVAPIKTPMIAGGLFVMDKFYFEELGKYDMMDVWGGENLEISFRVW PIKTP LAGGLF IDK YF EIG YD MDIWGGENLEISFRIW Consensus (301) G HSGALNAT1.pep (329)QCGGTLEIVTCSHVGHVFRKATPYTFPGGTGQIINKNNRRLAEVWMDEFK HSGAINAT2.pep (344)QCGGSLEIIPCSRVGHVFRKQHPYTFPGGSGTVFARNTRRAAEVWMDEYK Consensus (351)QCGGSLEII CS VGHVFRK PYTFPGGSG I KN RR AEVWMDEFK 450 401 HSGALNATI.pep (379)NFFYIISPGVTKVDYGDISSRVGLRHKLQCKPFSWYLENIYPDSQIPRHY HSGALNAT2.pep (394)NFYYAAVPSARNVPYGNIQSRLELRKKLSCKPFKWYLENVYPELRVPDHQ V YG I SRL LR KL CKPF WYLENIYPD IP H Consensus (401)NFFY 500 451 HSGAINAT1.pep (429) FSLGEIRNVEINQCLDNMARKENEKVGIFNCHGMGGNQVFSYTANKEIRT HSGALNAT2.pep (444) DIAFGALQQGIN-CLDTLGHFADGVVGVYECHNAGGNQEWALTKEKSVKH N TN CLD LA VGIF CH GGNQ FAT KIK Consensus (451) 501. HSGALNATI.pep (479) DDLCLDVSKLN--GPVIMLKCHHLKGNQLWEYDPVKLTIQHVNSNQCLDK HSGALNAT2.pep (493)MDLCLTVVDRAPGSLIKLQGCRENDSRQKWEQIEGNSKLRHVGSNLCLDS IL C Q WE L HV SN CLD Consensus (501) DLCL V 551 584 HSGAINAT1.pep (527) ATEEDSQVPSIRDCNGSRSQQWLLRNVTLPEIF-HSGALNAT2.pep (543) RTAK-SGGLSVEVCGPALSQQWKFTLNLQQ----S SI C A SQQW Consensus (551) T

FIG. 44 SUBSTITUTE SHEET (RULE 26)

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SUBSTITUTE SHEET (RULE 26)

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